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PRODUCT GUIDE

Neurodegenerative Diseases

upstate
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The Amyloid Protein Precursor (APP)

and Alzheimer's disease

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Alzheimer's disease is characterized by protein deposits, both inside and outside cells in the brain (7). The deposits found outside of cells, plaques, are comprised in their core of a peptide called the Aβ peptide. The Aβ peptide has been shown to be derived by cleavage of a large transmembrane precursor, the Alzheimer amyloid protein precursor (APP). Understanding the mechanisms and the causes of the cleavage of APP to release Aβ has been a primary focus in Alzheimer's research for the past 15 years. In addition, understanding the function of APP has been a similarly important focus.

While there was some historic controversy about whether Aβ levels are correlated with disease, it has now become quite clear that the levels of Aβ peptide inversely correlate with cognitive functioning in Alzheimer's disease [recent, large studies from the Mount Sinai brain bank include (14, 20, 21)]. One nuanced outcome of these and other studies was the demonstration that it is in fact the presence of the longer Aβ peptides (ending at amino acids 42 and 43) that best correlates with cognitive status in disease. These clinical findings are completely supported by genetic studies showing that very rare mutations in APP or in the presenilins lead to early-onset familial forms of Alzheimer's disease and to increased formation of longer Aβ peptides (22). Given the importance of the various subspecies of Aβ, much work has gone into developing immunological and other methods for precisely identifying and quantifying different Aβ species.

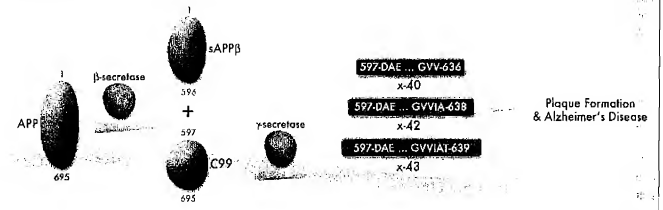
After the discovery of APP, the search for the enzymes responsible for the cleavage of APP became a major focus in Alzheimer's research. α-Secretases are responsible for cleaving APP at the COOH-terminus of Aβ. Overwhelming evidence implicates the presenilins as being directly involved in α-secretase activity (16). Presenilins are multipass transmembrane proteins that may function as novel aspartate proteases. Recent studies show that the presenilins function as a complex together with nicastrin, pen-2, and aph-1. This complex when reconstituted appears to have γ-secretase activity (15, 23). The presenilin/γ-secretase complex can cleave many proteins at or within the membrane (see figure on right). The enzyme responsible for cleaving at the NH2-terminal of the Aβ domain has been identified as BACE-1 (28). BACE-1 is a transmembrane proteases with a single transmembrane domain. Inhibitors of β- and γ-secretase activity are being studied as potential therapies in Alzheimer's disease (6, 19).

The first proteases to be discovered that cleave APP, with the exception of the signal peptidase which is responsible for the appropriate membrane insertion of APP, were actually the α-secretases. The α-secretases cleave APP within the Aβ domain and hence preclude the formation of Aβ (12). The α-secretases identified so far are transmembrane proteases with a single transmembrane domain [e.g., (1, 4, 18) for a review of the α-secretases]. The catalytic domain is in the extracellular

compartments, and recent studies have shown that over expression of α-secretase is protective in mouse models of Alzheimer's disease (24). A subset of the α-secretases can be activated by protein kinase C and other signal transduction cascades (2). The stimulation of α-secretase activity by activation of such pathways leads to decreased Aβ formation (3). Activators of protein kinase C are again protective in mouse models of Alzheimer's disease (10).

One of the most fascinating discoveries in signal transduction over the past several years (which arose to a large degree from the study of APP processing) has been the demonstration that many proteins including APP undergo a process called regulated intramembraneous proteolysis (RIP) (9). In this process, transmembrane protein are cleaved, very often by the presenilin/γ-secretase complex, in such a way that a cytoplasmic fragment is liberated. In the cases that have been studied, the cytoplasmic fragment can then either function within the cytoplasm or go to the nucleus and regulate transcription. The best characterized example of RIP is Notch, where it is known that the cleaved and released Notch intracellular domain (NICD) leads to transcriptional changes (27). A critical component of RIP with Notch is the interaction of the NICD with members of the CSL family of DNA-binding proteins. This interaction is required for the activity of the Notch intracellular domain as a regulator of transcription. As APP also undergoes RIP, it has become of great interest to determine whether there are other key components of the APP signaling machinery that are required for functional and/or transcriptional effects of APP. Numerous studies have implicated the FE65 family of proteins as being critical interactors with the APP intracellular domain (8, 11, 13). FE65 proteins not only modulate the localization of APP and the cleavage of APP with the subsequent formation of Aβ, but also mediate functions of APP and are critical for any transcriptional effects that might be mediated by APP (5, 17, 25, 26). Identifying the gene that may be regulated by APP/FE65 is currently one of the most exciting areas in Alzheimer's research.

APP/Aβ Pathway



The principle protein implicated in Alzheimer's disease is the transmembrane amyloid precursor protein (APP). Multiple APP isoforms, generated by alternative splicing, have been described with a 770 amino acid isoform being the largest and a 695 amino acid isoform being most prevalent in neuronal cells. Amyloid β (Aβ) is produced by sequential cleavage of APP by proteases called secretases. Proteolysis of APP by β-secretase, which cleaves APP695 after Met-596, produces a large soluble n-terminal fragment (sAPPβ) and a small membrane-bound c-terminal fragment (C99). The C99 fragment is further processed by γ-secretase cleavage within the trans-membrane region at Val-636, Ala-638 or Thr-639 to produce the three Aβ isoforms of 40, 42 or 43 amino acids, designated x-40, x-42 or x-43. The Aβ isoforms can induce neuronal apoptosis and aggregate into amyloid plaques. Progression of Alzheimer's disease is associated with increased levels of the x-42 and x-43 Aβ peptides.

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C. Harold and Leila Y. Mathers Research Professorship

After earning his PhD at the Weizmann Institute of Science in Israel, Dr. Buxbaum came to New York for a post-doctoral fellowship at Rockefeller University, with Dr. Paul Greengard. He then joined the faculty at Rockefeller, where he became an Assistant Professor and Associate Director of The Fisher Center for Alzheimer's Research. He joined the Mount Sinai faculty in 1997, and now holds the C. Harold and Leila Y. Mathers Research Professorship in the Departments of Psychiatry, Neurobiology and Genetics and Adult Development. Dr. Buxbaum is also Director of Molecular Genetics in the Seaver Autism Research Center. Dr. Buxbaum focuses on some of the most widespread and devastating psychiatric disorders, including Alzheimer's disease (AD), schizophrenia and autism.

APP/Aβ Products

Description	Cat. #	Price
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Anti-Amyloid Precursor Protein +1, Amy-5, CT	07-485	$259
Anti-Amyloid Precursor Protein +1, Amy-6, CT	07-486	$259
Anti-APP, Amyloid β peptide domain	07-592	$269
Anti-Amyloid β (Aβ)$_{x-40}$, clone 11A5-B10	05-799	$279
Anti-Amyloid β (Aβ)$_{x-42}$, clone 12F4	05-831	$259
Anti-Amyloid β (Aβ)$_{x-43}$, clone 9C4	05-798	$279